論文の内容の要旨

A study on CLE peptides as abiotic and biotic mediators

(非生物的及び生物的環境応答因子としての CLE ペプチドの研究)

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Plants sense environmental stimuli and convert them into cellular signals, which are transmitted to distinct cells and tissues to induce adequate responses. Plant hormones and small secretory peptides such as CLAVATA3/EMBRYO SURROUNDING REGION-RELATED (CLE) peptides often function as environmental stress mediators. In this study, I investigated whether and how CLE1–CLE7 peptides, which share the CLE domain with high sequence similarity, mediate environmental stimuli in *Arabidopsis thaliana*.

First, I investigated the expression of *CLE1–CLE7* genes upon various abiotic stimuli. *CLE1–CLE7* responded to different environmental stimuli, such as nitrogen deprivation, nitrogen replenishment, cold, salt and dark in a sophisticated manner (Chapter III-1). In addition, I found that dark treatment could induce *CLE2* and *CLE3* expression in roots (Chapter III-2). To further investigate *CLE2* and *CLE3* functions, I generated knockout mutants and estradiol inducible overexpressors. The phenotypic analysis revealed that *cle2* showed a chlorosis phenotype under light-deficient condition (Chapter III-2). This result suggests that *CLE2* was required for light-

deficient responses. Using the estradiol inducible CLE2 overexpressor, I found that induction of *CLE2* in roots induced the expression of various genes not only in roots but also in shoots, and genes related to light-dependent carbohydrate metabolism were particularly induced in shoots (Chapter III-3). These results together suggest that root-induced CLE2 functions systemically in light-dependent carbohydrate metabolism in shoots.

Second, I investigated whether *CLE1–CLE7* genes were responsive to biotic stimuli. *CLE3* showed significant responses upon immune elicitor flg22, Pep2, and immune hormone salicylic acid (SA) (Chapter III-4). Among *CLE1-CLE7*, only *CLE3* was up-regulated by SA in roots (Chapter III-4). Expression analysis of *CLE3* in the receptor mutants of SA, flg22 and Pep2 revealed that *CLE3* expression is regulated by NPR1-dependent SA signaling pathway, NPR1-dependent flg22-FLS2 signaling pathway, and NPR1-independent Pep2-PEPR1/2 signaling pathway (Chapter III-5). I further investigated the down-stream genes of *CLE3* by performing microarray analysis using its estradiol inducible overexpressor and found that root *CLE3* overexpression affected gene expression in both roots and shoots. In particular, defense related genes *ERF104, HSPRO2* and *ACBP3* were significantly up-regulated in shoots (Chapter III-6), suggesting that like CLE2, CLE3 may also function as a root-derived signal that regulates defense related gene expression in shoots. In *cle3* lines, I found that *ERF104, HSPRO2*, and defense markers, *WRKY33* and *PR1*, failed to appropriately respond to root-applied SA in shoots (Chapter III-7). In roots, SA responses of *ERF104*, *HSPRO2* and *PR1* were not affected in *cle3* mutants. These results suggest a possible role of root-derived CLE3 in regulation of systemic rather than local immune responses.

In conclusion, I found that CLE2 and CLE3 peptides, CLE domains of which differ by only a single amino acid, play distinctive roles in shade response and immune response, respectively. These findings provide a concept that, in addition to their functional redundancy, similar CLE1–CLE7 peptides may be involved in distinctive environmental responses to maintain the robust adaptive capacity in Arabidopsis plants.